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Renal Osteodistrophy: Histologic Evaluation After Renal Transplantation

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SUCCESSFUL RENAL transplantation (RT) is able to solve most problems of uremic or dialyzed patients (pt). However, renal osteodistrophy (RO) is often a long-lasting problem, and bone disease can even worsen after RT. In fact, significant changes in bone histology have been seen up to 5 years after the procedure in more than 80% of pt with a well-functioning kidney graft.[1] Several factors, including age,[2,3] sex,[4,5] underlying renal disease,[3] time of dialysis,[6] type and severity of RO pre-RT,[2,6] and also immunosuppressive drugs[4,7] may contribute to persistence or worsening of RO.

Therefore, bone disorders still are a relevant morbidity factor for RT, often difficult to handle. Dual-energy X-ray absorptiometry (DEXA) is useful to measure bone mineral density,[5] but mineralized bone biopsy (BB) is the gold standard to evaluate type and severity of RO. Only a few have prospectively evaluated bone histology after RT. We conducted a prospective study on renal transplant pt to analyze the histologic evolution of RO after RT.

PATIENTS AND METHODS

Eight pt who completed the study were submitted to a first BB on day 0 and a second BB 6 months after RT. The second BB was performed after 500 mg dimethylchlortetracycline/day, double labeling (3 days on, 10 days off, 3 days on). Transiliac bone biopsies using the trephine of Bordier-Meunier with an internal diameter of 8 mm were obtained in all pt. The bone specimens were fixed for 24 hours in methanol, dehydrated, and embedded in methylmetracrylate. Undecalcified sections of 5-μm thickness were made using a microtome (Model Leica RM 2155, Germany) equipped with a tungsten carbide knife. Three nonconsecutive sections were stained with Toluidine blue, acid phosphatase, and von Kossa. Two were stained with a specific histochemical stain for detection of aluminum in bone (azurin solochrom) and one with a PERLS histochemical stain for detection of iron in bone. Each bone section was read twice by two different persons, without knowledge of clinical or biochemical information on the pt. All sections were analyzed quantitatively for static parameters of bone formation and bone reabsorption. Histomorphometric measures were carried out using a semiautomatic image analyzer (Q-Win, Leica, Germany) coupled with a Leitz microscope. Approximately 40 different fields were analyzed for the same bone biopsy. Normal bone histomorphometric values are from Vernejoul et al.[8] The bone histomorphometric parameters below, expressed according to the standardized nomenclature,[9] were measured in trabecular bone.

All the pt were checked for intact parathyroid hormone (iPTH), total alkaline phosphatase (tAP), serum calcium (Ca), and serum phosphate (P) at time 0 and at 6 months. Serum creatinine was also checked at 6 month. No patient received calcium or calcitriol therapy after kidney transplantation. The immunosuppressive regimen used was cyclosporine A (CsA) and prednisone in all pt; one pt also had azathioprine; two pt also had antithymocytic globulin; and another two also had micophenolate mofetil. Steroids were used as three initial bolus of 500 mg methylprednisolone, followed by 0.5 mg/kg per day of prednisone, tapered to 10 mg/d at the end of the third month (our maintenance dose). None experienced acute rejection episodes, so no further steroid bolus were used.

For the statistical analysis, paired t test and Wilcoxon test were used.

RESULTS

Our study group was composed of five females and three males, with a mean age of 50.4 $\pm$ 9.8 years old (ranging from 27 to 57). Their mean time on dialysis (all of them were on hemodialysis) was 43.4 $\pm$ 31.3 months (5 to 97 months). The underlying renal disease was unknown in two pt, chronic glomerulonephritis in one pt, polycystic kidney disease in three pt, and chronic interstitial nephritis in two pt.

The mean and range of initial iPTH was 348.4 $\pm$ 440.1 pg/mL (38.3 to 1288), and the final iPTH was 76.9 $\pm$ 45.0 pg/mL (19 to 149), with the difference statistically significant ($P = .012$). The mean initial tAP was 75.5 $\pm$ 19.7 U/L, and the final tAP was 95.8 $\pm$ 32.8 U/L, although this increment was not significant ($P > .05$). There was a significant elevation on Ca during these 6 months (2.12 $\pm$ 0.16 vs 2.36 $\pm$ 0.13 mmol/L; $P = .001$) and a highly significant decrease on P between the two measurements (6.40 $\pm$ 0.95 vs 3.13 $\pm$ 0.24 mg/dL; $P < .001$). The mean serum creatinine at month 6 was 1.2 $\pm$ 0.3 mg/dL.

The histologic diagnosis from the initial and the final BB is shown on Table 1. Between the two BB, we verified an

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Table 1. Bone Histology and Their Evolution Between the Two BB

Patient	First BB	Second BB
1	Moderate HPTH; extensive Al deposits; normal mineralization	Mild HPTH; decrease in Al deposits; mineralization not significantly compromised
2	Mild HPTH, severe osteoporosis, extensive Al deposits	HPTH persists (predominant bone formation); Al deposits diminished; normal mineralization
3	Severe HPTH, without Al deposition; normal mineralization	Quite normal histology and normal bone volume
4	Moderate HPTH; significant Al deposits; normal mineralization	Improved HPTH, evolving to low-turnover bone disease; Al deposits decreased; osteoporosis
5	Quite normal bone histology; without significant Al deposition	Quite normal histology
6	Severe HPTH (predominant bone formation); extensive Al deposits; normal mineralization	Moderate HPTH; Al deposits decreased; mineralization mildly compromised
7	HPTH on the past, evolving to adynamic bone disease; extensive Al deposits; osteoporosis	Normal bone volume (osteoporosis improved); Al deposits diminished; normal mineralization
8	Low-turnover bone disease; normal bone volume; significant Al deposits	Mild HPTH; important decrease in Al deposits; normal bone volume

HPTH = hyperparathyroidism; Al = aluminum.

increase in mineralized bone, an increase in osteoblastic and osteoid surfaces, a decrease in osteoclastic surface and osteoclast number, and a decrease in aluminum surface. However, these changes were not statistically significant (see Table 2).

DISCUSSION

RT normalizes phosphate urinary excretion and calcitriol production,[2] two of the most important deficiencies of the uremic state implicated in RO. Also, aluminum overload is efficiently removed by a functioning RT, although this can take more than 1 year.[2] Dialysis-related amyloidosis complaints usually rapidly improve, although the cystic lesions persist.[2] However, severe pre-RT hyperparathyroidism (HPTH), especially nodular hyperplasia, may not improve after RT; indeed this may cause severe hypercalcemia and graft dysfunction,[10] making surgery necessary. Moreover, steroids given for RT have a well-known effect on bone, leading to bone mass loss[4] and contributing to avascular bone necrosis. CsA has also been associated with osteopenia[7] and with a disabling bone pain syndrome.[11] In addition, adynamic bone disease has increased in the last years,[3] and although its significance is uncertain, it may contribute to osteopenia observed after RT.

As easily understood from our results, some of the pt had severe aluminum load pre-RT, which expectedly decreased on second BB. However, high-turnover bone disease, although not extremely severe, was the most frequent cause of RO. These results are in accordance with the literature.[2,3] Pre-RT severe osteoporosis was also identified in two pt. Intact PTH and tAP decreased between the two measurements, as previously described,[12] and P showed the expected lowering. On the contrary, Ca significantly increased during this time interval, never reaching risky levels. This increment may be long lasting and probably is due to nonsolved HPTH.[10] All the pt always had a good graft function.

A significant loss in bone mass early after RT was made evident by DEXA measurements, predominantly at the lumbar spine. Up to 6.8%[12] or even up to 10%[2] reduction on bone mass in the first 5 or 6 months has been seen. Fortunately, this extremely high rate of bone loss progressively falls: there are studies showing a trend toward improvement after the sixth month[5] and recovery at month 12,[5] or normalization of bone mass only 2 years after RT.[2] The less encouraging results show that the loss still exists more than 8 years after RT, with a bone loss rate of 1.7% per year.[13]

However, there is a lack of studies with histologic analysis clarifying how the dynamic process of bone remodeling really is functioning after RT. Some of the few published works described a reduction in bone reabsorption, but with low bone formation rate, as early as 6 months[12] and even several months later.[14] Our study, with the handicap of only eight pt enrolled, showed a trend toward improvement in bone histology. Although the histologic changes between

Table 2. Bone Histomorphometry and Its Evolution Between the Two BB

	First BB	Second BB	*P* value
Mineralized surface (%)	32.99 ± 15.78	34.46 ± 11.08	.780
Osteoid surface (%)	4.11 ± 2.86	4.73 ± 2.68	.676
Trabecular surface (%)	37.10 ± 16.25	39.20 ± 12.02	.699
Osteoblastic surface (%)	5.72 ± 6.67	6.59 ± 5.46	.769
Osteoclastic surface (%)	3.05 ± 2.34	1.78 ± 1.87	.176
Trabecular surface with aluminum (%)	39.83 ± 35.38	30.98 ± 28.73	.228
Number of osteoclasts/mm^2	3.43 ± 2.79	2.45 ± 1.96	.302

the two BB did not reach statistical significance, we observed a trend toward improvement in bone formation and mineralization, with reduction of bone reabsorption and also important aluminum removal.

In conclusion, we can say that 6 months after RT there is already a trend toward improvement in RO, which can probably be clearly apparent a few months later, according to DEXA results. A longer follow-up is needed to confirm these results.

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